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TITLE: Immune-Stimulating Combinatorial Therapy for Prostate Cancer

PRINCIPAL INVESTIGATOR: Robert Ivkov

CONTRACTING ORGANIZATION: Johns Hopkins University
Baltimore, MD 21218

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13. SUPPLEMENTARY NOTES

14. ABSTRACT

In this project we aim to demonstrate a proof-of-concept in an animal model that combined radiation therapy and magnetic nanoparticle hyperthermia can elicit an anti-cancer immune response to inhibit progression of prostate cancer tumors. During this reporting period we 1) established a suitable tumor model and methods for inducing tumor immunologic effects with magnetic iron oxide nanoparticle (MION) hyperthermia and external beam radiation therapy; and, 2) developed methodologies that will be used to elucidate the role of key immune cell populations in tumors. Results obtained during this reporting period indicate that treatment of a model primary tumor by nanoparticle hyperthermia alone does not elicit a measurable response in a distal (untreated) tumor; whereas radiation therapy alone generated a modest response in distal tumors. Interestingly, the combination of radiation+hyperthermia produced the greatest observed distal tumor growth inhibition, and was associated with significantly elevated intratumor FOXP3, a master immune regulatory protein, levels. Radiation and hyperthermia single-agent therapies were associated with FOXP3 levels similar to untreated controls. These early results are encouraging and motivate further study.

15. SUBJECT TERMS
Hyperthermia, radiation therapy, immunotherapy, prostate cancer, magnetic nanoparticle(s), abscopal effect, immune cells, tumor-infiltrating immune cells, T-cells, CD4+/CD8+, cytokines, immune surveillance

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1. **INTRODUCTION:** The objectives in this effort are to demonstrate a proof-of-concept in a suitable animal model that combined ionizing (external beam) radiation and MION-mediated heating can elicit an immune response, and to elucidate mechanistic features of this response for future optimization of 'targeted' MION constructs. The specific objectives of efforts in this reporting period were to 1) establish a suitable tumor model and methods for inducing tumor immunologic effects with focal tumor heating from magnetic iron oxide nanoparticles (MIONs) and radiation; and, 2) assess methodologies that will be used to elucidate the potential role of key immune cell (e.g. CD4+, CD8+ T-cell) populations in tumors following these therapeutic interventions.
2. **KEYWORDS:** Hyperthermia, radiation therapy, immunotherapy, prostate cancer, magnetic nanoparticle(s), abscopal effect, immune cells, tumor-infiltrating immune cells, T-cells, CD4+/CD8+, cytokines, immune surveillance.

3. **ACCOMPLISHMENTS:**

- **What were the major goals of the project?**
  - Aim 1, Phase 1. Assess potential for tumor immune-modulating effects related to injection procedure and/or presence of MIONs.
  - Aim 1, Phase 2. Develop thermal dosimetry measured by inserted temperature probes, and assess effects of temperature probe presence.
  - Aim 1, Phase 3. Assess immune-modulating effects of dose-escalating HER2-MION HT.
  - Aim 2. Evaluate the immunologic response to RT+MION-HT +/- IT.
- **What was accomplished under these goals?**
  - Major activities in this effort included establishing a bilateral (primary-distal) tumor model and methods to assess systemic immune-modulating effects of therapeutic intervention(s) on the primary tumor with radiation therapy (RT), nanoparticle-mediated hyperthermia (HT), or the combination RT+HT +/- immunotherapy (IT). One over-arching objective of the current project is to demonstrate a robust *abscopal* effect, i.e. inhibition of growth of distal (untreated) tumor following treatment of the primary tumor, which is believed to result from immune-mediated signaling. The specific objective of activities in this first year was to establish tumor model and methods necessary to evaluate potential for immune-modulation with HT+RT. Given the objective requires an intact immune system in our animal (mouse) model, a mouse-derived prostate cancer cell line was necessary to enable use of immune-competent animal subjects. See below for summary animal numbers used.
  - Cell line: Mouse-derived prostate cancer cells, MyC-CaP, were obtained from the American Type Culture Collection (ATCC).
  - Mouse model: Male FVB/n (N=160) mice were used. Mice were ordered and used in smaller groups for focused methods development, testing, and training. Mice were housed for one week upon arrival before the experiment commenced to allow them to adjust after transport. MyC-CaP cells were cultured and ~1×10^6 cells suspended

in PBS were implanted subcutaneously into thighs of mice, and monitored with regular caliper measurements. Cells to generate a simulated 'primary' tumor were injected on the right thigh. Four days later, cells to generate a 'distal' tumor were injected on the left thigh to simulate metastatic or distal tumor site. After injection, and once the tumors were palpable, they were measured twice weekly with calipers. Growth of tumors in the bilateral

Mouse group	Number of mice	Group designation and Intended purpose	Notes
I	10	Pioneer, tumor growth implantation and assessment	Data were used to develop and modify procedures
II	10	Pioneer, MION injection, HT, thermal dosimetry	Data were used to develop and modify procedures
III	20	Therapy, C, HT, RT, RT+HT	Censored data were used (see figures)
IV	20	Therapy, C, HT, RT, RT+HT	Censored data were used (see figures)
V	20	Therapy, C, HT, RT, RT+HT	Censored data were used (see figures)
VI	20	Therapy, C, HT, RT, RT+HT	Cells were exposed to low CO2 during culturing. After implantation aberrant tumor growth was noted. Animal data were not used.
VII	30	Therapy, C, HT, RT, RT+HT	AMF equipment malfunctioned. Extended time for repairs was needed. Animals used for RT and C groups only (censored data).
VIII	30	Therapy, C, RT	Tumor volumes and tumor appearance was aberrant. Data were excluded.

model was studied to enable estimation of suitable time points for therapy. Tumor burden (i.e., tumor volume) is a significant factor for therapeutic outcomes, and it is also suspected to play a major role with immune suppression or inhibition of tumor progression. Implanted tumors, once established, grew rapidly demonstrating aggressive growth. At varying time points, tumors were harvested and tested for suitability to prepare histopathology samples and cell preparations for flow cytometry. Tumor growth, iron content and distribution analysis, tissue histopathology, and immune cell flow cytometry are principal endpoints of the study, thus preliminary activities in this phase were focused to establish reproducible methods. **Table 1** summarizes the breakdown of mice by groups, number, and purpose. **Figures 1** and **2** summarize the experimental flow and design that resulted from this major effort.

- Nanoparticles: In this study, bionized nanoferrite magnetic iron oxide nanoparticles (BNF-MIONs), an aqueous suspension of aggregated crystals of magnetic Fe_2O_3 /Fe_3O_4, were used.
- AMF Platform: The AMF system comprises three main components: the inductor coil, external capacitance network (Fluxtrol Inc., MI, USA), and an 80-kW power supply (PPECO, CA, USA). Together, the inductor coil (or inductor), external capacitance network and power supply form the resonant circuit. The AMF system was calibrated using a field probe (Fluxtrol Inc.), and field amplitude was measured in the coil center before each trial. During operation, the inductor coil and all AMF components were cooled using a closed-loop, circulating water system maintained between 22 and 25°C (Dry Cooler Systems Inc., MI, USA). In addition to minimizing

nonspecific heating from eddy currents, an additional circulating water shield, inserted within the inducting coil, was used to maintain a constant ambient temperature of 35-37°C during treatment.

- Small animal radiation research platform (SARRP): All treatments were performed using the SARRP, combining CBCT with ionizing radiation in a unified platform. Treatment was administered using a 3-mm focal spot with the X-ray tube maintained at 225 kVp. Radiation fields of 5 × 10 mm were used, ensuring coverage of the tumor while sparing other tissues and organs, for a 12-Gy total single-fraction dose.
- Therapy: Mice were anesthetized for injections (nanoparticle or saline), temperature probe insertion, and therapy (RT, HT and RT+HT) using isofluorane inhalation or ketamine/xylazine intraperitoneal injection, according to established and approved procedures. HT+RT were performed in the same treatment session under anesthesia. MIONs were percutaneously injected into the primary tumor. 24 hours after BNF-MION injection, mice were treated with alternating magnetic field (AMF) to generate heat in the primary tumor. Temperatures were measured at one-second intervals with AMF-resistant optical fiber temperature probes. Endpoints included tumor growth, iron quantification and histopathology. Mice were randomly assigned to one of four cohorts – no treat control (saline injection), radiation therapy (RT) (dose of 12 Gy, single fraction), nanoparticle hyperthermia (HT), single fraction target dose of CEM43 = 30-60 min, or RT+HT using same doses as in RT and HT cohorts.

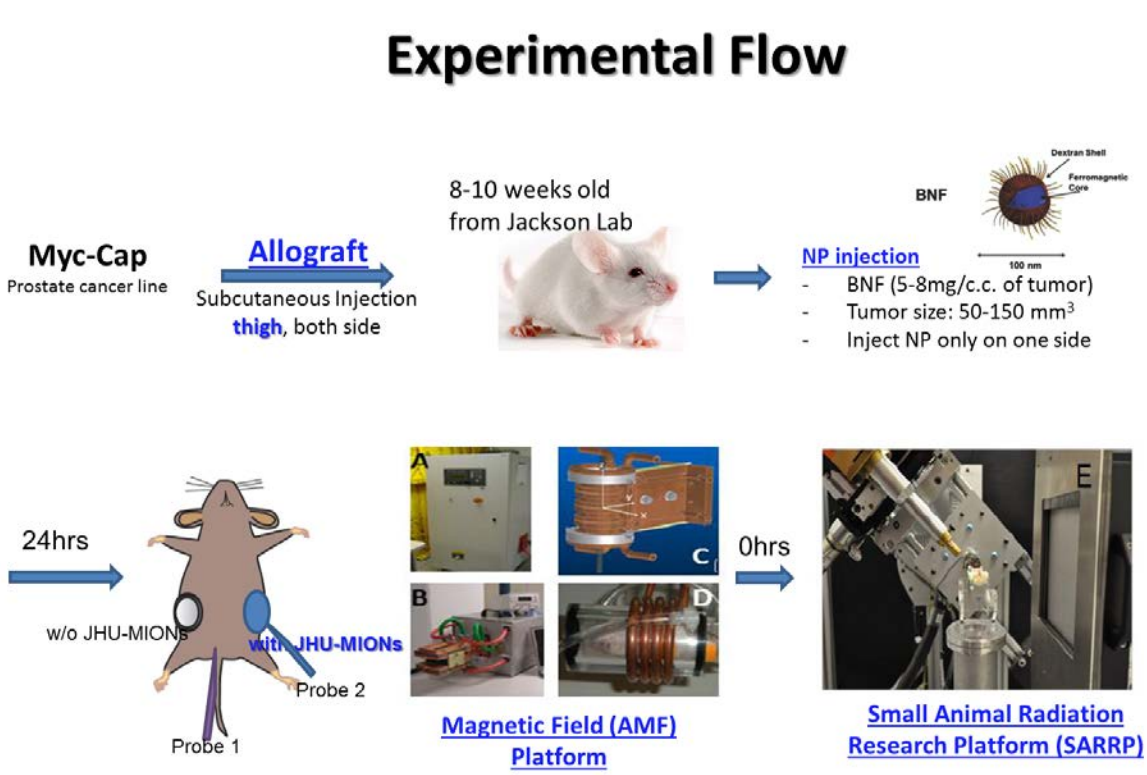

Figure 1: Schematic diagram of experimental flow established from major activities during this reporting period.

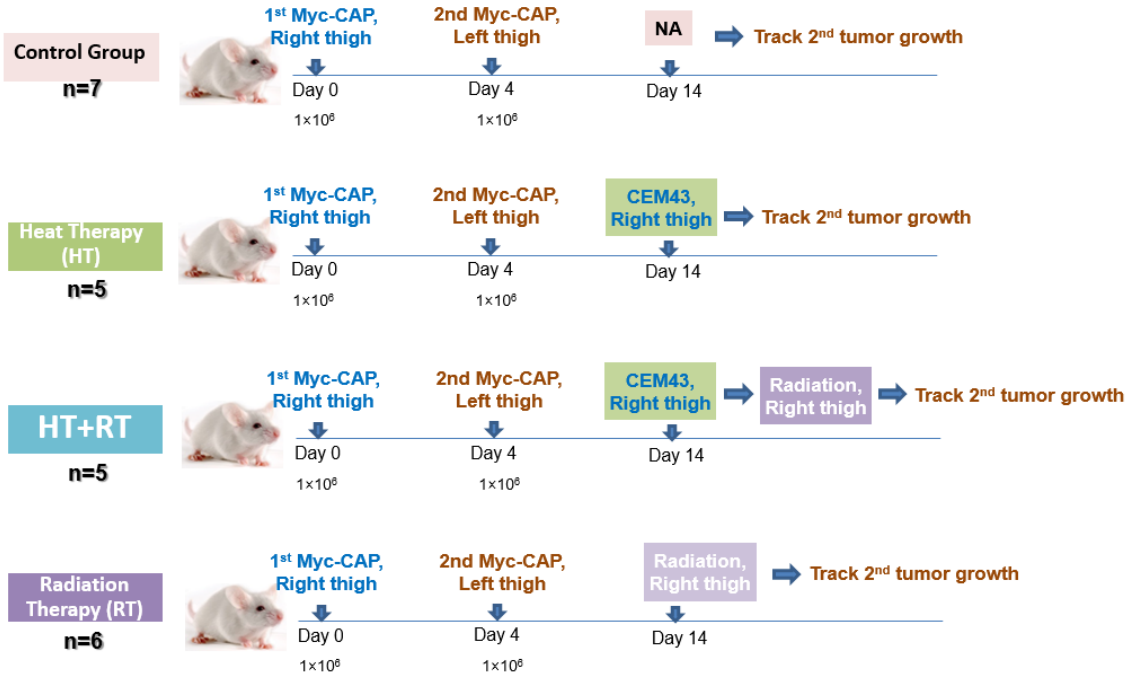

Figure 2: Flow chart of experimental design.

- Immune cell infiltrate analysis with flow cytometry: Tumors were resected at day 26, weighed and minced in RPMI with 10%FBS and filtered through 70µm strainers to harvest single cells. Red blood cells were lysed using ACK lysis buffer. Cells were then washed with media and lymphocytes were enriched by percoll gradient centrifugation. After centrifugation, the lymphocyte layer was separated and total number of live cells was counted. 2×10^6 cells were diluted in 100µl PBS for each tumor (except for RT+HT cohort primary tumor from which too few live cells were harvested) and incubated for 30 minutes at room temperature with fixable red dead stain for live/dead cells. After centrifugation and washing with PBS, cells were stained with fluorescently labelled CD4 and CD8 antibodies for 30 minutes at room temperature. After centrifugation and wash with PBS, cells were fixed and then stained with antiFoxP3 antibody for 20 minutes at room temperature. Cells were centrifuged and washed with PBS and then analyzed on a FACS calibur. Data analysis was performed using FlowJo software.

Specific accomplishments

- Aim 1, Phase 1. Assess potential for tumor immune-modulating effects related to injection procedure and/or presence of MIONs. Assessment(s) of injection procedure (saline control) was conducted. No discernible effect, as measured by tumor growth of distal (uninjected) tumor was observed (see **Figure 3**). Effect of MION injection and presence in tumor on tumor-infiltrating immune cells has not yet been completed.
- Aim 1, Phase 2. Develop thermal dosimetry measured by inserted temperature probes, and assess effects of temperature probe presence. Effects on distal tumor of temperature probe insertion was not tested during this reporting period; however, MION injection with AMF treatment at varying power levels to achieve CEM43 30-60 min was developed. Representative thermometry results are displayed in **Figure 4**.

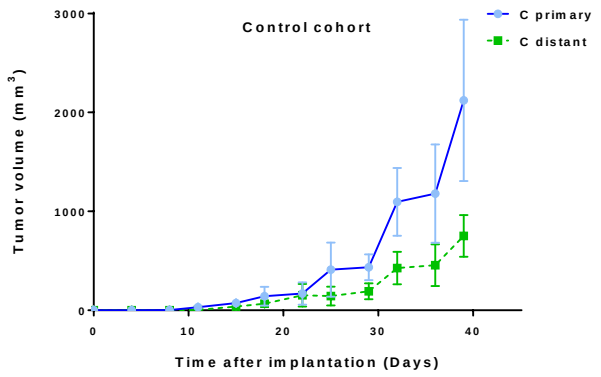

Figure 3: Tumor growth of control (primary injected with saline) cohort. Distal tumor was implanted four days after implantation of primary. Tumor growth rate of distal tracks that of primary, but its volume is slightly delayed due to later implantation. Results represent mean of n=15 individuals.

- Aim 1, Phase 3. Nothing to report
- Aim 2. Evaluate the immunologic response to RT+MION-HT +/- IT. Analysis of tumor growth data demonstrates that HT, RT, and HT+RT affect primary tumor growth, compared to control (C). RT (12 Gy) and RT+HT display a more pronounced growth inhibiting effect on the primary tumor than does HT alone (**Figure 5A**). By comparison, distal tumor growth was inhibited by RT or RT+HT, compared to control, with the latter demonstrating a slightly more pronounced effect (**Figure 5B**). Distal tumor growth following HT was not inhibited, and the current results suggest there may be a slight acceleration of growth. While these preliminary results are tantalizing, the small

number of subjects (n<5) is not statistically robust and mandates caution. We noted evidence of the dependence

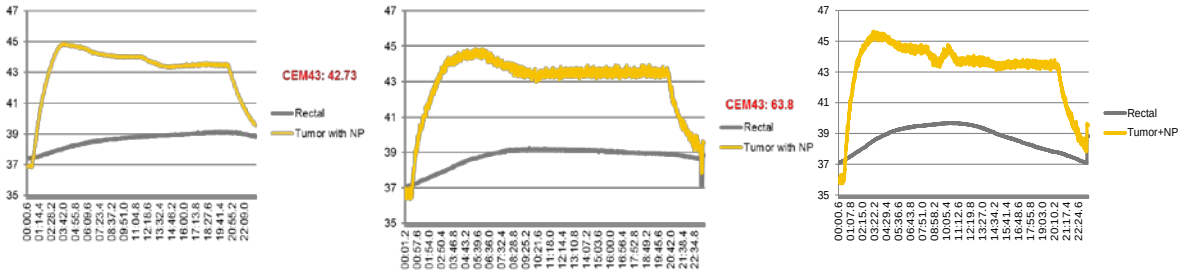

Figure 4: Heat therapy delivered within targeted dose of CEM43 30-60 min. Representative thermal graphs of hyperthermia therapy with MIONs. Yellow trace shows measured intratumor temperatures, and black traces are rectal temperatures.

of treatment outcome on initial volume of both primary and distal tumors at time of treatment, in a preliminary analysis of data (not shown). Large or small primary (or distal) tumors at time of treatment produced generally unresponsive distal tumor growth to HT, RT, or RT+HT. Data in **Figures 5** summarize results of retrospectively censored data in which primary tumor at time of treatment fell within a medium range of 50 – 100 mm^3 (0.05-0.1 cm^3), thus reducing total numbers of subjects used in the analysis. In a separate 'prospective' study with RT alone, we compared results of tumor growth following RT only, supporting the observation that volume of the primary tumor at the time of RT treatment influences growth of distal tumor (data not shown). The primary tumors

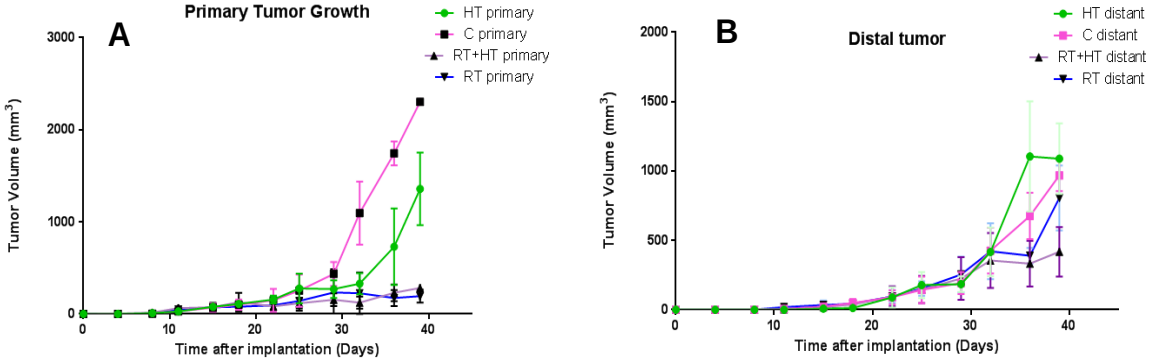

Figure 5: Measured volumes of primary (A) and distal (B) tumors with time following treatment of primary tumor only, with one of saline control (C, n=6), single fraction of ionizing radiation with dose 12 Gy (RT, n=4), single fraction of hyperthermia with dose of CEM43 = 30-60 min (HT, n=3), and combined RT+HT (n=3). Data presented were censored retrospectively to include subjects having tumor volumes in the range primary = 50 – 100 mm^3 and distal = 0 – 50 mm^3 at time of treatment.

were categorized into small (0-0.05 cm^3), medium (0.05-0.1 cm^3), and large (0.1-0.25 cm^3) size at the time of treatment. All primary tumors across all sizes demonstrated growth inhibition compared to control. The medium sized primary tumors had the most suppressed growth overall. For the distal tumors, those having the medium size at time of treatment also had suppressed growth, however due to their initial size, the relative growths varied significantly.

- Results of immune flow cytometry yielded similarly intriguing results. Primary tumor response to RT was evident, by increased CD4+ and CD8+ T-cell populations; however there was relatively little evidence of changed T-cell infiltration following either HT or RT+HT relative to controls in distal tumors. The notable exception is the markedly increased FoxP3 protein expression in distal tumors following combined RT+HT. This finding merits further study.

	CD4	CD8	FoxP3
Control Pri-T	2.5	0.99	3.28
Control Dis-T	1.47	0.75	8.17
HT- Pri-T	2.41	1.38	6.94
HT-Dis-T	2.01	1.13	8.62
RT-Pri-T	4.17	3.4	6.25
RT-Dis-T	1.77	1.42	0
HRT-Pri-T	-	-	-
HRT-Dis-T	1.99	1.2	15.5

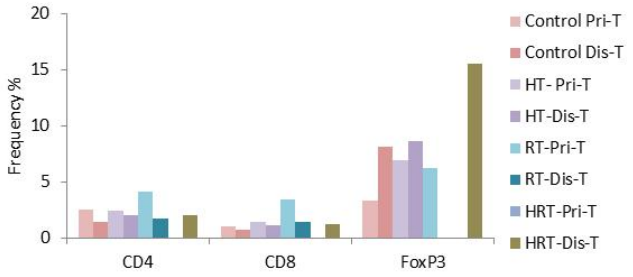

Figure 6: Flow cytometry analysis of tumor-infiltrating cells shows slightly increased inflitration of CD4+ and CD8+ cells into primary tumors following radiation therapy (RT-Pri-T), as compared to either control (Control Pri-T) or HT (HT-Pri-T); however, the presence of T-cell infiltrates in distal tumors following RT (RT-Dis-T) is relatively similar to that of both control and HT treated groups. Interestingly, FoxP3 expression is markedly increased in distal tumors of RT+HT (HRT-Dis-T) cohorts.
Key: Control- PBS; HT- Hyperthermia alone; RT- Radiation alone; HRT- Hyperthermia and Radiation; Pri-T- Primary tumor; Dis-T- Distal tumor

- Aim 1, Phase 1. Assess potential for tumor immune-modulating effects related to injection procedure and/or presence of MIONs. Our preliminary assessment revealed no discernible effects on distal tumor growth following saline injection, however these were not compared against “no treat” control groups, and there was not a separate comparison with nanoparticle injections. There has been some recently published evidence that nanoparticles can interact with tumor infiltrating macrophages to induce primary tumor growth inhibition when co-injected with cells. While this specific experiment is outside the scope of the current effort, the published results highlight the importance to ascertain within our model whether the MIONs present in primary tumors may alter the population or behavior of tumor-infiltrating T-cells.
- Aim 1, Phase 2. Develop thermal dosimetry measured by inserted temperature probes, and assess effects of temperature probe presence. Additional replicate experiments are needed to improve statistical significance of results.
- Aim 1, Phase 3. Assess immune-modulating effects of dose-escalating HER2-MION HT. These studies will commence in the coming reporting period.
- Aim 2. Evaluate the immunologic response to RT+MION-HT +/- IT. As above, additional experiments are needed to establish statistically robust data for analysis and interpretation. Additional samples for histopathology evaluation, iron content/distribution analysis, and immune-cell flow cytometry is needed.

- **What opportunities for training and professional development has the project provided?**

- **Training provided by this project:**
    - **Dr. Shu-Han Yu (post-doctoral fellow)**
        - Flow cytometry training on FACS caliber. Training provided by JHU SOM Core Facility personnel.
        - Fundamentals of Cancer: Cause to Cure, (Course), JHU SOM Course, two weekly lectures during Fall Semester.
        - Animal Exposure Surveillance Program (AESP), JHU.
    - **Dr. Preethi Korangath (Research Associate)**
        - Flow cytometry training on FACS caliber. Training provided by JHU SOM Core Facility personnel.
        - Fundamentals of Cancer: Cause to Cure, (Course), JHU SOM Course, two weekly lectures during Fall Semester.
    - **Jacqueline Stewart (Technologist)**
        - Flow cytometry training on FACS caliber. Training provided by JHU SOM Core Facility personnel.
        - Rodent surgery, JHU IACUC.
- **Professional development provided by this project:**
    - **Dr. Shu-Han Yu (post-doctoral fellow)**
        - Attendance at several seminars (weekly) – Assistant Professor Summer Lecture Series; Translational Research Con.
        - Fundamentals of Cancer: Cause to Cure, (Course), JHU SOM Course, two weekly lectures during Fall Semester.
        - Animal Exposure Surveillance Program (AESP), JHU.
    - **Dr. Preethi Korangath (Research Associate)**
        - Attendance at several seminars (weekly) – Assistant Professor Summer Lecture Series; Translational Research Conference; GU Oncology Departmental seminars (weekly) – all at JHU SOM.
        - Fundamentals of Cancer: Cause to Cure, (Course), JHU SOM Course, two weekly lectures during Fall Semester.
    - **Jacqueline Stewart (Technologist)**
        - Flow cytometry training on FACS caliber. Training provided by JHU SOM Core Facility personnel.

- Rodent surgery, JHU IACUC.

- **Also please include any and all conferences, workshops, training seminars, etc.**
- **How were the results disseminated to communities of interest?**
    - **Dr. Shu-Han Yu (post-doctoral fellow)**
        - Presentation of results (poster) at International Congress for Hyperthermic Oncology, April, New Orleans, LA.
        - Presentation of results (seminar) at MRS 2016 Series Progress Seminar, March, Baltimore, MD.
        - Presentation of results (poster) at Radiation Oncology Scientific Retreat, 2016, February, Baltimore, MD.
- **What do you plan to do during the next reporting period to accomplish the goals?**
    - During the next reporting period, we will further optimize methodology and data analysis to identify and separate "responders" from "non-responders" to therapy. We will assess the potential for immune cell modulation of the presence of nanoparticles in tumors. We will commence with immunotherapy combinations. And, we will implement injection of our MION-HER2 constructs in MyC-CaP-HER2 expressing tumors.

4. **IMPACT:**

- **What was the impact on the development of the principal discipline(s) of the project?**
    - Nothing to report
- **What was the impact on other disciplines?**
    - Nothing to report.
- **What was the impact on technology transfer?**
    - Nothing to report
- **What was the impact on society beyond science and technology?**
    - Nothing to report.

5. **CHANGES/PROBLEMS:**

- **Changes in approach and reasons for change**
    - For the present effort, and moving forward we have implemented a transition to MyC-CaP (murine prostate cancer) tumor model. The TRAMP model has proven challenging with unpredictable yields in breeding (TRAMP mice are inefficient breeders) producing too few offspring that would generate sufficient numbers of autochthonous tumors. It has proven to be an unreliable and inferior model for purposes of this study. In addition, we have available a HER2-expressing variant of the MyC-CaP

cells, providing the appropriate model system to test immune cell sensitization and immunotherapy. The direction, methods, and goals of the study remain unchanged.

- **Actual or anticipated problems or delays and actions or plans to resolve them**
  - As described above. The changes and plans have been developed (see above material) and implemented.
- **Changes that had a significant impact on expenditures**
  - Although there has been no significant impact on expenditures for this reporting cycle, Dr. Drake (co-investigator) will now collaborate from Columbia University Medical Center. A request for approval to initiate a subaward will be submitted to the USAMRAA Grants Officer. Dr. Drake is critical to the success of this project. No subaward will be issued to Columbia University until USAMRAA approval is received.
- **Significant changes in use or care of human subjects, vertebrate animals, biohazards, and/or select agents**
  - Nothing to report
- **Significant changes in use or care of human subjects**
- Nothing to report
- **Significant changes in use or care of vertebrate animals.**
- MyC-CaP model has replaced TRAMP?
- **Significant changes in use of biohazards and/or select agents**
- Nothing to report

6. **PRODUCTS:**

- **Publications, conference papers, and presentations**
  - **Journal publications.** Nothing to report
  - **Books or other non-periodical, one-time publications.** Nothing to report.
  - **Other publications, conference papers, and presentations.**
    - **<u>Dr. Shu-Han Yu (post-doctoral fellow)</u>**
      - Presentation of results (poster) at International Congress for Hyperthermic Oncology, April, New Orleans, LA.
      - Presentation of results (seminar) at MRS 2016 Series Progress Seminar, March, Baltimore, MD.
      - Presentation of results (poster) at Radiation Oncology Scientific Retreat, 2016, February, Baltimore, MD.

- **Website(s) or other Internet site(s)**

  Nothing to report

- **Technologies or techniques**

  Nothing to report

- **Inventions, patent applications, and/or licenses**

  Nothing to report

- **Other Products**

  Nothing to report

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

- **What individuals have worked on the project?**

Name:	***Robert Ivkov***
Project Role:	*Principal Investigator*
Researcher Identifier (e.g. ORCID ID):	
Nearest person month worked:	*2 calendar months*
Contribution to Project:	*He oversees all aspects of the proposed work and the testing conducted at Johns Hopkins*
Funding Support:	*No other funding support for this project. See Other Support document for comprehensive, detailed funding support.*
Name:	***Shu-Han Yu***
Researcher Identifier (e.g. ORCID ID):	
Nearest person month worked:	*4 calendar months*
Project Role:	*Post-doctoral Fellow*
Contribution to Project:	*A post-doctoral (PhD) biology/bioengineering with focus in immunology. She assisted with design and execution of experiments and performed analysis (flow cytometry, temperature, ICP-MS, etc.) under the general supervision of the PI. She participated in all aspects of planning experiments and*

	presentation of results.
Name:	***Jacqueline Stewart***
Researcher Identifier (e.g. ORCID ID):	
Nearest person month worked:	*4 calendar months*
Project Role:	*Research Technologist*
Contribution to Project:	*The laboratory technician is supervised by Dr. Ivkov. She performs cell harvest and maintains the cells (for adoptive transfer, or analysis, etc.), monitors animals, collects blood and tissue samples, and conducts injections, and assists with therapy and imaging experiments, performs mass spectrometry and sample preparation, and serves as technical assistant to the postdoctoral fellow.*
Funding Support:	*No other funding support for this project.*

- **Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?**
  - *Please refer to Other Support documents for details.*
- **What other organizations were involved as partners?**

  Nothing to Report

8. **SPECIAL REPORTING REQUIREMENTS**
   - **COLLABORATIVE AWARDS:** Nothing to report.
   - **QUAD CHARTS:** Nothing to report.
9. **APPENDICES:** N/A

OTHER SUPPORT

IVKOV, ROBERT

Changes to Active support since last submission: NIH grant R01CA194574 was funded and effort on the Jayne Koskinas Ted Giovanis Foundation was reduced from 50% to 25%.

Active Support

Title: **Nanoparticles and their targeting in preclinical models**
Time Commitment: 25% effort (3 Calendar Months)
Sponsor: Jayne Koskinas Ted Giovanis Foundation
Name of Procuring Contracting/Grants Officer: Theodore Giovanis
Address of Funding Agency: 7141 Deer Valley Road, PO Box 130, Highland, MD 20777
Period of Performance: 01/01/15-12/31/17
Level of Funding: $335,000 Annual Direct Costs
Project Goal: To develop targeted nanoparticle constructs that will provide robust platforms for cancer imaging and therapy.
Specific Aims:

1) Characterize tumor structure and nanoparticle uptake, distribution and potential for therapy of a HER2-targeted nanoparticle construct in subcuataneous human breast cancer models.
2) Characterize tumor structure and nanoparticle uptake and distribution in immune-competent mice bearing HER2+ implanted tumors.
3) Perform pilot optimization of anti-HER2 directed therapy using HER2-targeted nanoparticle hyperthermia to achieve durable response in transgenic HER2+ mice that develop spontaneous tumors with metastatic progression.

Project's overlap or parallel: No scientific or budgetary overlap
Role: PI

Title: **Nanotechnology 2020: Preparing students to use nanotechnologies to solve contemporary problems in agriculture and human sciences**
Time Commitment: 15% effort (1.8 Calendar Months)
Sponsor: University of Arizona
Name of Procuring Contracting/Grants Officer: Gricelda La Turco
Address of Funding Agency: 888 N. Euclid Ave, Room 515, Tucson, AZ 85719
Period of Performance: 12/01/14-11/30/17
Level of Funding: $39,480 Annual Direct Costs
Project Goal: To develop online course modules (nanomedicine and characterization focus), and increase interdisciplinary collaboration with partner institutions.
Specific Aims:

1) Prepare course modules in online format.
2) Participate in pilot testing with consortium.
3) Aid implementation of courses at JHU.

Project's overlap or parallel: No scientific or budgetary overlap
Role: PI

Title: **Enhancing Liver Cancer Treatment with Image-Guided Magnetic Hyperthermia**
Time Commitments: 28% effort (3.4 Calendar Months)
Supporting Agency: NCI R01CA194574 (Liapi, Ivkov Co-PI)
Grants Officer: Jacquelyn Saval, 240-276-6312, boudjedaj@mail.nih.gov

Address of Funding Agency: National Cancer Institute, Executive Plaza South, Suite 243, 6120 Executive Blvd., Bethesda, MD 20892-7150
Performance Period: 05/01/2015-04/30/2020
Level of Funding: $410,164 annual direct costs ($3,245,481 total costs)
Project's Goal: The goal of this project is to determine if x-ray image-guided magnetic hyperthermia chemosensitization for treatment of unresectable primary liver cancer leads to superior treatment effects when compared to standard-of-care chemotherapy.
Specific Aims:

1) Assess local and systemic distribution using x-ray image-guided injection of MIONs in a large animal model of liver cancer
2) Develop an imaging methodology to concurrently assess MION intra-tumor deposition and tumor perfusion changes following MHT
3) Assess treatment efficacy of combined image-guided MHT and chemotherapy in xenograft mouse models of liver cancer

Project's overlap: No scientific or budgetary overlap with current proposal
Role: Co-Principal Investigator

Title: **PC140189 Immune-Stimulating Combinational Therapy for Prostate Cancer**
Time Commitments: 20% effort (2.4 Calendar Months)
Supporting Agency: CDMRP
Grants Officer: Kimberly Carter
Address of Funding Agency: W03J USA Research Mat CMD, 1077 Patchel Street, Fort Dietrick, MD 21702-5024
Performance Period: 09/30/15-09/29/18
Level of Funding: $125,000 annual direct costs ($607,500 total costs)
Project's Goal: The goal of this project is to provide preclinical data to motivate development of a targeted MION for future development of a prostate cancer vaccine.
Specific Aims:

1) Assess the effects and mechanisms of HT (single treatment) on development of immunologic responses.
2) Determine effects of HT+RT +/- IT on immunologic responses.

Project's overlap: No scientific or budgetary overlap with current proposal
Role: Principal Investigator

Pending Support: N/A

OTHER SUPPORT

DRAKE, CHARLES G.

Changes to Active support since last submission: Dr. Drake is now faculty at Columbia University Medical Center. His effort on the following projects will transfer with appropriate Sponsor approval.

Active Support

R01CA154555 (Drake)
Title: Role of Tc17 cells in tumor immunotherapy
Effort: 2.28 calendar months (19% effort)
Supporting Agency: National Cancer Institute
Name of Procuring Contracting/Grants Officer: Connie Murphy
Address of Funding Agency: 6120 Executive Blvd, EPS/Suite 243, Rockville, Md. 20892-7150
Performance Period: 03/01/12- 02/28/2017
Level of Funding: $207,5000 annual direct cost
Project's Goal: These studies have broad clinical and immunological significance: successful completion of this work could transform adoptive T cell transfer for the treatment of cancer patients, and shed novel insight into fundamental aspects of CD8 function and differentiation.
Specific Aims: 1.) Define the cytokine and cellular requirements for Tc17 mediated immunotherapy in vivo 2.) Understand the TCR/peptide and peptide/MHC interactions critical for Tc17 skewing in vitro
3. **)**Establish the requirements for Tc17 conversion to an IFN-γ secreting phenotype 4.) Determine the molecular mechanisms underlying Tc17 persistence in vivo.
Role: PI
Overlap: None

90055855 (Drake)
Title: Comprehensive Transcriptional Profiling of Human Prostate-cancer infiltrating cells
Effort: .12 calendar months (1% effort)
Supporting Agency: Janssen Research and Development LLC
Name of Procuring Contracting/Grants Officer: Joseph Erhardt
Address of Funding Agency: 920 Route 202 South, Raritan, NJ, 08869
Performance Period: 09/09/2013-01/08/2017
Level of Funding: $206,100 annual direct
Project's Goal: The major goals of this project are to establish a specific immunologic profile of prostate cancer and identify new potential immunological targets to combat Tcell exhaustion and ultimately improve outcomes for patients with prostate cancer by allowing for discovery of specific immunologic therapies for prostate cancer that will create a durable immune response
Specific Aims: 1.) Create an immunologic profile unique to prostate infiltrating lymphocytes as compared to matched peripheral blood lymphocytes by compating naïve activated Tcells to determine which receptors are associated with exhaustion versus activation in CD4+ and CD8+ lymphocytes. 2.) Evaluate immunologic phenotype of surrounding epithelial cells of the tumor microenvironment as compared with that of adjacent normal tissue to identify potential molecular tumor targets as well as co-inhibitory immunological receptors

Role: PI
Overlap: None

SU2C-AACR-DT10 (Pardoll)
Title: Immune Checkpoint Blockade and Adoptive Cell Transfer in Cancer Therapy
Effort: .24 calendar months (2% effort)
Supporting Agency: University of Texas M.D. Anderson Cancer Center (AACR Prime)
Name of Procuring Contracting/Grants Officer: Renee Gonzales
Address of Funding Agency: 1515 Holcombe Blvd, Houston, Texas 77030
Performance Period: 03/01/2013-02/28/2017
Level of Funding: $468,182 annual direct costs
Project's Goal: The major goal of this project is to enable the rapid and rational clinical investigation of new discoveries in one of the most promising areas of oncology research today, immune checkpoint blockade.
Specific Aims: 1.) Interrogation of immune responses within the tumor microenvironment before and after treatment with immune checkpoint blockade 2.) Interrogation of the targets of T and B cell responses after checkpoint blockade 3.) Development of combinatorial cancer therapies based on checkpoint blockade.
Role: Co-Investigator
Overlap: None

90054364 (Pardoll)
Title: International Immuno-Oncology Network-IION Resource Model
Effort: .12 calendar months (1% effort)
Supporting Agency: Bristol-Myers Squibb Co
Name of Procuring Contracting/Grants Officer: Les Enterline
Address of Funding Agency: Route 206 & Province Line Road, Princeton, NJ 08543
Performance Period: 05/07/2013-05/06/2017
Level of Funding: $486,987 annual direct costs
Project's Goal: The major goal of this clinical research network is to conduct immunotherapy trials with novel agents including anti-KIR, anti-CD137 and others, and to collaboratively evaluate pharmacodynamics and potential biomarkers of response.
Specific Aims: 1.) Analyze immune-inhibitory networks in resected tumors employing 3 techniques for geographic localization: (i) IHC, (ii) amplified ISH, and (iii) qRT-PCR analysis of laser capture micro-dissected (LCM) regions of leukocytic infiltration. 2. Complementary to the studies in 1, we will sort myeloid, lymphoid and cancer cells from freshly dissociated tumors in cases where enough tumor is available, allowing analysis by flow cytometry and mRNA profiling of cellular subsets for co-expression of inhibitory ligands, receptors and druggable metabolic enzymes.
Role: Co-Investigator
Overlap: None

CA224-020 (Drake)
Title: A Phase 1 Dose Escalation and Cohort Expansion Study of the Safety, Tolerability, and Efficacy of Anti-LAG-3 Monoclonal Antibody (BMS-986016) Administered Alone and in

Combination with Anti-PD-1 Monoclonal Antibody (Nivolumab, BMS-936558) in Advanced Solid Tumors
Effort: .12 calendar months (1% effort)
Supporting Agency: Bristol Myers Squibb Co
Name of Procuring Contracting/Grants Officer: Dan Fontana
Address of Funding Agency: Route 206 & Province Line Road, Princeton, NJ 08543
Performance Period: 11/12/2013-11/11/2017
Level of Funding: $911,969 total direct costs
Project's Goal: The goal of this clinical trial is teo study the safety, tolerability, and efficacy of Anti-LAG-3 monocloal antibody (BMS-986016) administered along and in Combination with Anti-PD-1 Monoclonal Antibody (Nivolumab, BMS-936558) in Advanced Solid Tumors
Specific Aims: N/A
Role: PI
Overlap: None

90061378 (Drake)
Title: Understanding Checkpoint Expression and Function in GBM RCC and Bladder CA by Integrated Analysis of Tumor Infiltrating Lymphocytes and Tumor Cells
Effort: .12 calendar months (1% effort)
Supporting Agency: Bristol-Myers Squibb Co
Name of Procuring Contracting/Grants Officer: Les Enterline
Address of Funding Agency: Route 206 & Province Lane Road, Princeton, NJ 08543
Performance Period: 11/19/2014-04/06/2017
Level of Funding: $141,453 annual direct costs
Project's Goal: The major goals of this project is to determine the relative expression of known and novel checkpoint molecules in pathologist-curated patient samples and the functional significance of these molecules using micro-scale functional assays.
Specific Aims: N/A
Role: PI
Overlap: None

GO29313 (Drake)
Title: A Phase 1, Open-Label, Dose-Escalation Study of The Safety and Pharmacokinetics of MOXR0916 Administered Intravenously As a Single Agent to Patients with Locally Advanced or Metestatic Solid Tumors
Effort: .12 calendar months (1% effort)
Supporting Agency: Genentech Corporation
Name of Procuring Contracting/Grants Officer: Wayne Athers
Address of Funding Agency: 1 DNA Way South, San Francisco, CA 94080
Performance Period: 07/07/2014-12/08/2017
Level of Funding: $2,721,282
Project's Goal: The major goal of this trial is to evaluate the safety and tolerability of MOXR0916 in patients with locally advanced or metastatic tumors
Specific Aims: N/A
Role: PI
Overlap: None

90061946 (Drake)
Title: Epigenetic Drugs and Immuno Therapy for Prostate Cancer (EDIT-PC)
Effort: 1.2 calendar months (10% effort)
Supporting Agency: Prostate Cancer Foundation
Name of Procuring Contracting/Grants Officer: Howard R. Soule, PhD
Address of Funding Agency: 1250 Fourth Street, Santa Monica, CA 90401
Performance Period: 12/24/2014-12/23/2017
Level of Funding: $210,000 annual direct costs
Project's Goal: To evaluate the ability of a novel, multivalent cancer vaccine based on attenuated listeria monocytohenes (*Lm)* to induce prostate cancer-specific immune responses, and to attenuate tumor progression
Specific Aims: 1.) Evaluate a novel, trivalent prostate cancer vaccine based on an attenuated listeria platform for safety, tolerability and preliminary evidence of efficacy in men with metastatic castration-resistant prostate cancer (mCRPC). 2.) Determine the magnitude and breadth of antigen-specific T and B cell immune responses induced by this novel vaccine. 3.) Using biopsies of metastatic lesions, quantify the induction of a pro-inflammatory immune infiltrate as well as expression of checkpoint ligands (including PD-L1) for potential utility as predictors of response and/or resistance.
Role: PI
Overlap: None

15003789 (Paller)
Title: Overcoming drug resistance in metastatic castration resistant prostate cancerActivation of Specific
Effort: .6 calendar months (5% effort)
Supporting Agency: The Community Foundation for the National Capital Region
Name of Procuring Contracting/Grants Officer: K. Matthews
Address of Funding Agency: 1201 15th St, NW, Suite 420, Washington, DC 20005
Performance Period: 11/14/2014-11/13/2019
Level of Funding: $884,956 total direct costs
Project's Goal: The goal of this clinical trial is to evaluate a new combination therapy to extend the life of men with advanced prostate cancer.
Specific Aims: N/A
Role: Co-Investigator
Overlap: None

90061256 (Pardoll)
Title: Analysis of Novel Immunodulatory Ligands and Receptors
Effort: .12 calendar months (1% effort)
Supporting Agency: Compugen Ltd.
Name of Procuring Contracting/Grants Officer: Anat Cohen Dayag, President & CEO
Address of Funding Agency: Pinchas Rosen Street #72, Telaviv 69512, Israel
Performance period: 12/17/2014-11/30/2019
Level of Funding: $331,395 annual direct costs

Project's Goal: The major goal of this project is to study the immunobiology and cancer immunotherapy relevance of multiple novel gene products identified as potentially immunomodulatory.
Specific Aims: N/A
Role: Co-Investigator
Overlap: None

W81XWH-15-1-0667 (Ivkov)
Title: Immune-Stimulating Combinatorial Therapy for Prostate Cancer
Effort: 0.6 calendar months (5% effort)
Supporting Agency: Department of Defense CDMRP
Grants Officer: Kathy E. Robinson
Address of Funding Agency: US Army Medical Research & Materiel Command, 820 Chandler Street, Fort Detrick, MD 21702-5014
Performance Period: 09/30/15-09/29/18
Level of Funding: $125,000 annual direct costs
Project's Goal: The goals of this project are to 1) induce tumor immunologic effects with focal tumor heating from magnetic iron oxide nanoparticles (MIONs) and radiation; and, 2) assess role of cytokines (e.g. ILs) and key immune cell (e.g. CD4+, CD8+ T-cell) populations in tumors to assess immune response(s) to HT and HT+RT +/- IT.
Specific Aims: 1) Assess the effects and mechanisms of HT (single treatment) on development of immunologic responses. 2) Determine effects of HT+RT +/- IT on immunologic responses.
Role: Co-Inv
Overlap: None

90065447 (Drake)
Title: The Effects of Nivolumab on the T Cell Phenotype and Tumor Microenvironment in Patients with Resectable RCC
Effort: .12 calendar months (1% effort)
Supporting Agency: Bristol Myers Squibb Co
Grants Officer: Rahbar H Tayyabkhan
Address of Funding Agency : Route 206 & Province Lane Road, Princeton, NJ 08543
Performance Period: 09/24/2015-03/23/2017
Level of Funding: $332,441annual direct costs
Project's Goal: The primary endpoint of this study will be safety / feasibility, given that all patients will undergo a pre-enrollment biopsy and subsequent surgical resection, we will have the ability to perform comprehensive biomarker studies using both treated and untreated tissues.
Specific Aims: 1) Quantify the effects of nivolumab monotherapy on RCC TIL in humans. 2)Understand the effects of nivolumab monotherapy on the stromal / myeloid components of the TME 3) Correlate baseline cytokine profiles (and changes in cytokine profile) with nivolumab-driven CD8 infiltration.
4) Test whether TCR clonality in the PBL of tumor correlates with induced CD8 infiltration.
Role: PI
Overlap: None

W81XWH-15-1-0670 (PI: Nelkin/Drake/Luo)
Title: CDK5-A Novel Role in Prostate Cancer Immunotherapy
Time commitment: .96 calendar months (8% effort)
Supporting agency: CDMRP
Procuring Contracting/Grants Officer: Kathy Robinson
Address of Grants Officer: 820 Chandler Street, Fort Detrick, MD
Performance period: 09/30/2015-03/29/2018
Level of funding: $150,000
Project's Goal(s): The goal of this project is to develop a novel, effective targeted therapeutic strategy for advanced prostate cancer, blocking several of the most common resistance mechanisms to androgen deprivation therapy (ADT), that underlie progression to castration resistant prostate cancer (CRPC)
Specific Aims: 1. Effect of dinaciclib on androgen receptor (AR) S81 phosphorylation and function. **2**. Effect of dinaciclib, alone and in combination with inhibitors of potential compensatory signaling pathways, in human prostate cancer cell lines and xenografts. **3**. Effect of dinaciclib combinations in a model of prostate cancer bone metastasis.
Role: MPI
Overlap: None

(PI:Drake)
Title: Understanding PD1 Function in RCC by Analyzing Extremes of Response - A Biomarker Study
Time commitment: .30 calendar months (2.5% effort)
Supporting agency: Bristol Myers Squibb Co
Procuring Contracting/Grants Officer: Monique R. Adams, PhD
Address of Grants Officer: Route 206 & Province Line Road, Princeton, NJ 08543
Performance period: 03/29/2016-03/28/2017
Level of funding: $16,235 annual direct costs
Project's Goal(s): The goal of this project is to analyze a bioinformatic study of pre-existing data from RCC patients, according to according to response group (extreme responders versus extreme progressors) will yield predictive and/or on-study biomarkers and will further identify key features of response leading to the initiation of next-generation clinical trials
Specific Aims: 1. Cytokine analysis: test the hypothesis that either pre-treatment cytokine levels, or on-treatment changes in cytokine levels will correlate with response group. **2.** Microarray analysis test the hypothesis that the baseline transcriptional signature from tumor biopsies will correlate with response group. We will also test the alternative hypothesis that on-treatment changes in transcripts correlate with response group **3.** Microarray analysis: We will test the hypothesis that the baseline transcriptional signature from tumor biopsies will correlate with response group. We will also test the alternative hypothesis that on-treatment changes in transcripts correlate with response group.
Role: PI
Project Overlap or Parallel: No scientific or budgetary overlap.

(PI: Pardoll)
Title: The Johns Hopkins University Bloomberg-Kimmel Institute for Cancer Immunology
Time commitment: 2.4 calendar months (20% effort)

Supporting agency: Bloomberg Philanthropies
Procuring Contracting/Grants Officer: Patricia Harris
Address of Grants Officer: 25 E. 78th St, New York, NY 10075
Performance period: 09/30/2015-03/29/2018
Level of funding: $10,000,000
Project's Goal(s): The goal of the Institute is to develop, within 10 years, immunotherapies that can place 50% of people with inoperable cancer into lifelong remission.
Specific Aims: N/A
Role: Associate Director, GU Program Leader and Co-Leader of the Immunomodulation Program
Overlap: None

Pending Support:

R01CA214879 (Pienta)
Title: Tumor promoting macrophages as a therapeutic target for metastatic prostate cancer
Effort: 1.2 calendar months (10% effort)
Supporting Agency: NIH/NCI
Name of Procuring Contracting/Grants Officer: TBD
Address of Funding Agency: 6120 Executive Blvd, Suite 243 Rockville, MD 20892
Performance Period: 04/01/2017-03/31/2022
Level of Funding: $499,973 annual direct costs
Project's Goal: This proposal will delineate the roles of M2-tumor associated macrophages (M2-TAMs) in promoting prostate cancer (PCa) tumorigenesis, dissect the dynamic relationship between M2-TAMs and host immune cells in preclinical models and clinical specimens, and develop a unique and effective targeting strategy against M2-TAMs. The proposed studies take original and important steps towards understanding the contribution of M2-TAMs to tumorigenesis and developing novel treatments for metastatic PCa.
Specific Aims: 1: Delineate the roles of M2-TAMs in promoting PCa tumorigenesis. **2:** Dissect the dynamic relationship between M2-TAMs and host immune cells inpreclinical models and clinical specimens. **3:** Target M2-TAMs for prostate cancer therapy.
Role: Co-Investigator
Overlap: None

OTHER SUPPORT

DEMARZO, ANGELO

Changes to Active support since last submission: Dr. DeMarzo's effort on the following awards has ended since the last report: Dr. DeMarzo's Johns Hopkins Internal Award, Dr. Laiho's Prostate Cancer Foundation (90059710), Dr. Sfanos' Johns Hopkins Internal Award; Dr. Yegnasubramanian's Johns Hopkins Internal Award; Dr. Farokhzad's Prostate Cancer Foundation award; Dr. Lotan's W81XWH-12-PCRP-TIA; Dr. Platz' W81XWH-12-1-0545 & W81XWH-12-1-0170; Dr. Drake's Prostate Cancer Foundation; and Dr. Trock's Prostate Cancer Foundation Movember award.
New Awards since last reporting period: Dr. Luo's W81WXH-14-PCRP-BDA PC141019; Dr. Trock's & DeMarzo's PC140318; and Dr. Zhang's U01CA152813

Active Support

R01CA185297
Title: The aberrant androgen receptor underlies abiraterone/enzalutamide resistance
Effort: 0.24 calendar months (2%)
Supporting Agency: NIH/NCI R01CA185297
Grants Officer: Jacquelyn Saval, 240-276-6312, boudjedaj@mail.nih.gov
Address of Funding Agency: National Cancer Institute, Executive Plaza South, Suite 243, 6120 Executive Blvd., Bethesda, MD 20892-7150
PI: Luo J/ Antonarakis
Role: Co-Investigator: DeMarzo
Performance Period: 05/01/15-04/30/19
Level of Funding: $228,750
Description of Goals: The goal of this project is to dissect molecular drivers of therapeutic resistance by validating the AR-V concept in the clinical setting of men receiving abiraterone and enzalutamide for metastatic prostate cancer, and to ultimately overcome AR-V-mediated therapeutic resistance.
Specific Aims:
Aim 1: To determine whether positive detection of prostate cancer-derived AR splice variant-7 (AR-V7) is associated with primary or acquired resistance to abiraterone and enzalutamide.
Aim 2: To define the transcriptional landscape of the aberrant AR by using RNA-Seq analysis.
Aim 3: To determine the role of AR-FL and AR-V dimer formation in mediating aberrant AR signaling after the canonical AR axis is rendered inactive through potent AR-directed therapies.
Projects overlap or parallel: No scientific or budgetary overlap.

W81WXH-14-PCRP-BDA (Luo)
PC141019
Title: Non-invasive detection of AR-FL/AR-V7 as a predictive biomarker for therapeutic resistance in men with metastatic castration-resistant prostate cancer
Time Commitment: 0.48 calendar months (4%)
Supporting Agency: US Department of Defense
Name of Procuring Contracting/Grants Officer: Kathy E. Robinson
Address of Funding Agency: 820 Chandler St., Fort Detrick, MD 21702-5014
Performance Period: 09/30/15-09/29/18
Level of Funding: $347,125
Project Goal: Our overall objective is to develop indicators or predictors of therapeutic response and resistance to abiraterone and enzalutamide by focusing on mRNA-based tests compatible with serial non-invasive blood sampling in men with metastatic CRPC.
Specific Aims:

1: To perform cross-institutional analytical validation of a blood-based assay in a certified environment (CLIA or international equivalent).
2: To expand existing prospective clinical correlation studies to enable assay qualification and clinical validation.
3: To plan, coordinate, and facilitate multi-institutional clinical trials integrating AR biomarkers.
Projects overlap or parallel: There is no scientific or budgetary overlap.

W81XWH-15-1-0667 (Ivkov)
Title: PC140189 Immune-Stimulating Combinatorial Therapy for Prostate Cancer
Time Commitment: 0.21 Calendar Months (1.75%)
Supporting Agency: Department of Defense CDMRP
Grants Officer: Kathy E. Robinson
Address of Funding Agency: US Army Medical Research & Materiel Command, 820 Chandler Street, Fort Detrick, MD 21702-5014
Performance Period: 09/30/15-09/29/16
Level of Funding: $125,000 annual direct costs
Project's Goal: The goals of this project are to 1) induce tumor immunologic effects with focal tumor heating from magnetic iron oxide nanoparticles (MIONs) and radiation; and, 2) assess role of cytokines (e.g. ILs) and key immune cell (e.g. CD4+, CD8+ T-cell) populations in tumors to assess immune response(s) to HT and HT+RT +/- IT.
Specific Aims:
1- Assess the effects and mechanisms of HT (single treatment) on development of immunologic responses.
2- Determine effects of HT+RT +/- IT on immunologic responses.
Project's overlap: No scientific or budgetary overlap with current proposal
Role: Co-Inv

P50CA58236 (De Marzo, PI of Core B)
Title: SPORE in Prostate Cancer, Core B – Specimen Core
Effort: 0.96 calendar (8%)
Supporting Agency: US National Institutes of Health/ US National Cancer Institute
Grants Officer: NIH/NCI Grants Associate- AMO
Address of Funding Agency: NIH 616 Executive Boulevard, Suite 7013, MSC 8347, Rockville, MD 20852/ NCI Public Inquires Office 6116 Executive Boulevard Room 3036A Bethesda, MD 20892-8322
Performance Period: 9/25/14 - 08/31/19
Level of Funding: $123,284
Project's Goals: The major goals of this Core are to maintain and enhance a repository of prostate tissues containing a wide range of neoplastic and non-neoplastic samples from both fresh frozen and paraffin blocks, prostatic fluids, DNA, RNA, and protein, to formalize standard workflows, operating and quality control policies and procedures for the collection, storage to distribute these samples to SPORE and other investigators as needed, and to perform innovative biospecimen research using these specimens.
Specific Aims:
1) To maintain and enhance a repository of prostate tissues containing a wide range of neoplastic and non-neoplastic samples from both fresh frozen and paraffin blocks, prostatic fluids, DNA, RNA, and protein, and, to distribute these samples to SPORE and other investigators as needed
2) To provide high quality histopathologic diagnoses of tissue specimens and tissue microarrays.
3) To perform well-controlled immunohistochemistry (IHC) assays, interpretation and quantitative analyses of IHC slides to facilitate the achievement of the specific aims of the individual research projects.
4) To continue to design, produce and distribute tissue microarrays using human prostate tissues, cell lines, and xenografts.
5) To continue to improve and add tools to our open source tissue microarray database and software (TMAJ) (http://tmaj.pathology.jhmi.edu) including the development and dissemination of new open source image

analysis tools "FRIDA" (FRamework for Image Dataset Analysis) while ensuring compatibility with the CaBIG grid system.
6) To test and potentially implement emerging software tools, such as CaTissue, from the CaBIG program for specimen banking efforts, serving as a model for other SPOREs and other research programs throughout our University.
7) To provide a facility and pathology expertise for laser capture microdissection.
8) To continue to function as the Central Pathology Core for the Inter-Prostate SPORE Biomarker Study (IPBS).
Projects overlap or parallel: No scientific or budgetary overlap

W81XWH-14-2-0182 (Trock PI; De Marzo, Co-PI)
Title: PC131930 Prostate Cancer Biorepository Network (PCBN) Johns Hopkins Prostate Cancer Pathology Resource Network Coordinating Center
Effort: 0.60 calendar months (5%)
Supporting Agency: DOD
Grants Officer: Nrusinghas C. Mishra, Ph.D.
Address of Funding Agency: DOD Prostate Cancer Research Program, CDMRP, U.S. Army Medical Research and Materiel Command MCMR-CD, 1077 Patchel Street, Fort Detrick MD 21702-5024; telephone: 301-619-7782; fax: 301-619-7796
Performance Period: 09/30/14-09/29/17
Level of Funding: $201,312 direct/yr
Project Goal: This project will coordinate among four institutions the development of a prostate cancer bio-repository with well-annotated specimens obtained using optimized and standardized protocols, and to conduct biospecimen science to characterize critical factors influencing the molecular integrity of research tissues.
Specific Aims:
1. Review of sources of patients and biospecimens at site
2. Participate in development of draft SOPs, common consent formats, and MTA.
3. Participate in SOP training.
4. Enroll patients and collect specimens.

Projects overlap or parallel: No scientific or budgetary overlap

W81XWH-14-2-0182 (De Marzo, Network Site PI)
Title: PC131930 Prostate Cancer Biorepository Network (PCBN) Resource Network Site for the Prostate Cancer Pathology Resource Network Award
Effort: 1.2 calendar months (10%)
Supporting Agency: US Department of Defense
Grants Officer: Nrusinghas C. Mishra, Ph.D D
Address of Funding Agency: DOD Prostate Cancer Research Program, CDMRP, U.S. Army Medical Research and Materiel Command MCMR-CD, 1077 Patchel Street, Fort Detrick MD 21702-5024; telephone: 301-619-7782; fax: 301-619-7796
Performance Period: 09/30/14-09/29/17
Level of Funding: $318,482 direct/yr
Project's Goal/Specific Aims: The goal of the PCPRN and the JHCC is to develop a biorepository with high quality, well-annotated specimens obtained in a systematic, reproducible fashion using optimized and standardized protocols, and an infrastructure to facilitate the growth of the resource and its wide usage by the prostate cancer research community. The PCPRN will also conduct and support biospecimen science that characterizes critical factors influencing the molecular integrity of research tissues.
Projects overlap or parallel: No scientific or budgetary overlap

90058581
Myriad Genetics Inc. (PI: Trock**)**
Title: Biomarker Predication of Metastatic Progression

Effort: 0.60 calendar months (5%)
Sponsor: Myriad Genetics Inc.
Grants Officer: Steven Stone
Performance Period: 03/01/14-02/28/17
Address of Funding Agency: 1077 Patchel Street Fort Detrick MD 21702-5024
Level of Funding: $358,421/yr
Project Goals: The goal of this study is to evaluate the performance of PTEN and a novel cell cycle based RNA profile for predicting outcomes in men who do or do not develop metastatic prostate cancer after radical prostatectomy, as well as for predicting which men are candidates for salvage radiation and hormonal therapy after prostatectomy.
Projects overlap or parallel: No scientific or budgetary overlap

R01CA190430 (Wheelan S/Yegnasubramanian S)
Title: Epigenetic Control of Retrotransposons in Human Cancers
Effort: 0.12 calendar months (1%) (no salary support)
Supporting Agency: NIH/NCI
Grants Officer: Paul Okano
Address of Funding Agency: NIH 6116 Executive Boulevard, Suite 7013, MSC 8347, Rockville, MD 20852
Performance Period: 09/01/14-08/31/19
Level of Funding: $250,000
Project's Goal: The goal of this project is to understand the epigenetic regulation of L1 retrotransposition in normal and cancer cells.
Specific Aims:
Aim 1: Use our computational and experimental systems to determine the extent to which DNA hypomethylation at cis regulatory elements near full length active L1 elements can lead to their transcriptional activation in cancer cells.
Aim 2: Identify the impact of DNA methylation changes in controlling L1 retrotransposition rate and genomic target site preference.
Aim 3: Determine the cis correlation of DNA hypomethylation with L1 transcription and with sites of L1 retrotransposition in human cancer tissues.
Projects overlap or parallel: No scientific or budgetary overlap

90048977
PCF Young Investigator Award (Joshu)
Title: Biological pathways underlying weight gain as a cause of prostate cancer recurrence
Time Commitment: 0.48 calendar months (4.03%)
Name and Address of the Funding Agency's Procuring Contracting/Grants Officer: Howard Soule 1250 Fourth Street Santa Monica, California 90401
Supporting Agency: Prostate Cancer Foundation
Performance Period: 02/01/12-01/31/15 - no cost extension until 11/16/16
Level of Funding: No cost extension
Project Goals: The goal of the project is to investigate two candidates' biological mechanisms that may explain our previous observation that weight gain is associated with prostate cancer recurrence: telomere length and inflammation.
Specific Aims:
1) To evaluate whether weight gain and obesity are associated with short telomere length in cancer-associated stromal cells and variable telomere length in cancer cells among men surgically treated for clinically localized prostate cancer.
2) To evaluate whether weight gain and obesity are associated the extent of inflammation present in benign and malignant prostate tissue among men surgically treated for clinically localized prostate cancer.
Projects overlap or parallel: No scientific or budgetary overlap

W81XWH-14-1-0364 CDMRP (Sfanos)
Title: Infections and Innate Immunity in Prostate Cancer Racial Disparities-PC132011
Sponsor: US Army Research Council
Name of Procuring Contracting/Grants Officer: Theresa Miller
Address of Funding Agency: 1077 Patchel Street Fort Detrick MD 21702-5024
Performance Period: 09/30/14-09/29/17
Level of Funding: $237,471/ direct/yr
Principal Investigator: Karen Sfanos
Effort: 1.2 calendar months (10%)
Project Goals: The goals of this project are to study differences in inflammatory markers in African American versus Caucasian American prostate cancer patients.
Specific Aims: 1. Quantification of mast cells and macrophages in prostate cancer and matched benign tissues from AA and CA patients. 2. Evaluation of the expression pattern and cellular localization of IL-1β, IL-6, IL-8, and IL-10 in both low grade and high grade prostate cancer tissues from AA and CA patients. 3. Assessment of infections agents present in prostate tissues from AA and CA patients using Illumina sequencing.
Projects overlap or parallel: No scientific or budgetary overlap

PC140318P1
Title: MYC RNAi-PT Combination Nanotherapy for Metastatic Prostate Cancer Treatment
Time Commitment: 0.60 Calendar Months (5%)
Supporting Agency: Department of Defense CDMRP (DeMarzo – PI)
Grants Officer: N/A
Address of Funding Agency: US Army Medical Research & Materiel Command, 820 Chandler Street, Fort Detrick, MD 21702-5014
Performance Period: 09/30/15-08/31/18
Level of Funding: $62,500 Annual Direct Costs
Project's Goal: The main objective of this project is to develop an innovative nanotherapy modality by combining platinum (Pt) chemotherapy and MYC-targeting RNA interference (RNAi) for more effective treatment of metastatic prostate cancer (PCa).
Specific Aims:
1: Development and optimization of MYC siRNA-Pt NPs. This aim includes three major tasks: (1) rational design and creation of a library of NPs for both siRNA delivery and Pt agent encapsulation; (2) evaluation of in vitro cellular cytotoxicity of these NPs in Pt-naïve and resistant PCa cells; and (3) in vivo test (e.g., pharmacokinetics, biodistribution, and toxicities) and optimization of the MYC siRNA-Pt NP system in PCa xenograft models.
2: Determination of the efficacy of select NPs in the B13MYC/Cre|Ptenfl/fl engineered PCa mouse model. We will (1) evaluate the NP biodistribution and MYC silencing in this mouse model, and (2) investigate tumor development and progression to metastasis after NP administration, as well as in vivo side effects.
Project's overlap: No scientific or budgetary overlap with current proposal

Title: U01CA152813 Glycoprotein biomarkers for the early detection of aggressive prostate cancer
Time Commitment: 0.24 Calendar Months (2%)
Supporting Agency: NIH (PI – Zhang)
Grants Officer: Jacquelyn Saval, 240-276-6312, boudjedaj@mail.nih.gov
Address of Funding Agency: National Cancer Institute, Executive Plaza South, Suite 243, 6120 Executive Blvd., Bethesda, MD 20892-7150
Performance Period: 09/01/15-03/31/21
Level of Funding: $359,999 annual direct costs
Project's Goal: This is a biomarker developmental laboratory (BDL) of the Early Detection Research Network (EDRN). The goal of this project is to identify glycoprotein biomarkers for the early detection of aggressive prostate cancer in tissues and urine.
Project's overlap: No scientific or budgetary overlap with current proposal

OTHER SUPPORT

DEWEESE, THEODORE

Changes to Active Support since last submission:

Dr. DeWeese is contributing 5% effort on Dr. Tran's Prostate Cancer Foundation grant; Grant U01CA183031 was funded; Grant R01CA161613 ended on 8/31/15; Grant R01CA151395 ended on 04/30/16; Dr. DeWeese's Advantagene trial ended; Grant P30CA006973 was added with 6% effort for Dr. DeWeese.

Active Support:

(Tran) Altering the Natural History of Metastatic Prostate Cancer using Stereotactic Ablative Radiotherapy and Immune Stimulation

Time Commitment: 5% effort (0.6 Calendar Months)
Supporting Agency: Prostate Cancer Foundation
Grants Officer: Audrey Gardner, Manager of Program Administration, agardner@pcf.org
Address of Funding Agency: Prostate Cancer Foundation, 1250 4th Street, Santa Monica, CA 90401; 310.570.4792
Performance Period: 08/01/15-07/31/17
Level of Funding: $500,000 annual direct costs
Project Goal: The goal of this project is to test the importance of treating all sites of disease with SABR in combination with the immune stimulatory agent ADXS-PSA in men with oligometastatic PCa to leverage this concept to full advantage for men suffering from metastatic PCa.
Specific Aims: 1. To examine circulating tumor cells (CTCs), circulating tumor DNA (ctDNA) and T-cell receptor (TCR) repertoire profiling as biomarkers for men with oligometastatic prostate cancer treated with stereotactic ablative radiation therapy (SABR) alone.
2. To conduct a first-in-man trial of stereotactic ablative radiation therapy (SABR) in combination with the immune stimulatory agent ADXS-PSA for men with oligometastatic hormone sensitive prostate cancer (HSPC).
3. To conduct a Phase II trial of stereotactic ablative radiation therapy (SABR) in combination with the immune stimulatory agent ADXS-PSA for men with oligometastatic prostate cancer.
Overlap: No scientific or budgetary overlap.

U01CA183031 (Pomper/DeWeese) PSMA Directed Imaging of Prostate Cancer Focus on Androgen Receptor Dynamics

Time Commitment: 10% effort (1.2 Calendar Months)
Supporting Agency: NIH/NCI
Grants Officer: Name of Procuring Grants Officer: Jacquelyn Saval, boudjedaj@mail.nih.gov, 240-276-6312
Address of Funding Agency: 9000 Rockville Pike; Bethesda, Maryland 20892
Performance Period: 07/14/15-06/30/18
Level of Funding: $245,595 annual direct costs
Project Goal: The goal of this project is to validate DCFPyL clinically so that it can be used to full advantage in supporting existing and emerging therapies for a spectrum of patients suffering from PCa.
Specific Aims: 1. To compare the performance of DCFPyL vs. DCFBC for imaging patients with biopsy-proven PCa and evidence of new or progressive metastatic disease by conventional imaging (CT and/or bone scan).
2. Image treatment-naïve patients with localized-locally advanced primary PCa using DCFPyLPET/magnetic resonance imaging, and correlate signal with that on MR concurrently obtained, as well as with tumor grade, PSMA expression and androgen receptor (AR) signaling before and after two months of neoadjuvant androgen deprivation (ADT).
3. Image patients with CRPC using DCFPyL-PET/MR and correlate with bone and soft tissue biopsy.

4. Image patients with CRPC with DCFPyL-PET/MR and correlate with standard 99mTc-based bone scan to guide stereotactic body radiation treatment (SBRT) in patients with oligometastatic disease.

Overlap: No scientific or budgetary overlap

90062232 (DeWeese) Improving Radiotherapy Conditions in Greece

Time Commitment: 1% effort (0.12 Calendar Months)
Supporting Agency: Stavros S Niarchos Foundation
Grants Officer: Programs Department, +1-212-486-7475, PRG-SNFNY@snf.org
Address of Funding Agency: 86A Vasilissis Sofias Avenue, 11528 Athens, Greece +30-210-877-8300
Performance Period: 10/17/14-01/31/17
Level of Funding: $250,000 total project costs
Project Goal: On-site assessment to improve radiation therapy technology and clinical care in Greece.
Specific Aims: N/A
Overlap: No scientific or budgetary overlap

90058751 (Wong/DeWeese) Exhibit F-1 Oncospace: An eScience Program for the Advancement of Care in Oncology

Time Commitment: 2% effort (0.24 Calendar Months)
Supporting Agency: Toshiba Corp.
Grants Officer: Masahiro Ozaki, +81-287-26-6210, masahiro.ozaki@toshiba.co.jp
Address of Funding Agency: 1385, Shimoishgami, Otawara-shi, Tochigi-ken 324-8550, Japan
Performance Period: 02/01/14-01/31/17
Level of Funding: $397,061 total project costs
Project Goal: The goal of this project is to expand the development of the Oncospace Program to: a) extract and combine imaging and non-imaging metadata forming a patient profile from clinical practice for a large population of patients in an oncology database; b) carry out data analysis on combined data to discover correlations that are clinically significant; c) support and inform treatment decisions; d) demonstrate potential for improving patient outcomes.
Specific Aims: 1. Implement multi-modality image processing tools for target definition, automatic feature extraction and other quantitative imaging metrics as meta-data.
2. Investigate the relationship of imaging metrics and non-imaging data with treatment outcomes for treatment optimization.

Overlap: No scientific or budgetary overlap.

W81XWH-15-1-0067 (Ivkov) PC140189 Immune-Stimulating Combinatorial Therapy for Prostate Cancer

Time Commitment: 1% (.12 Calendar Months)
Supporting Agency: Department of Defense CDMRP
Grants Officer: Kimberly Carter, 301-619-2249, Kimberly.m.carter47.civ@mail.mil
Address of Funding Agency: US Army Medical Research & Materiel Command, 820 Chandler Street, Fort Detrick, MD 21702-5014
Performance Period: 09/30/15-09/29/18
Level of Funding: $125,000 annual direct costs
Project's Goal: The goals of this project are to 1) induce tumor immunologic effects with focal tumor heating from magnetic iron oxide nanoparticles (MIONs) and radiation; and, 2) assess role of cytokines (e.g. ILs) and key immune cell (e.g. CD4+, CD8+ T-cell) populations in tumors to assess immune response(s) to HT and HT+RT +/- IT.
Specific Aims: 1. Assess the effects and mechanisms of HT (single treatment) on development of immunologic responses.
2. Determine effects of HT+RT +/- IT on immunologic responses.

Project's overlap: No scientific or budgetary overlap with current proposal

Role: Consultant

P30CA006973 (Nelson) Regional Oncology Research Center
Time Commitment: 7% (.9 Calendar Months)
Supporting Agency: NCI
Grants Officer: Jacquelyn Saval, boudjedaj@mail.nih.gov, 240-276-6312
Address of Funding Agency: 9000 Rockville Pike; Bethesda, Maryland 20892
Performance Period: 05/07/97-04/30/17
Level of Funding: $4,318,066 annual direct costs
Project's Goal: The major goal of this project is to provide an organizational focus and stimulus to take maximum collective advantage of scientific opportunity and institutional resources aimed toward the ultimate goal of reducing cancer incidence, morbidity, and mortality.
Project's overlap: No scientific or budgetary overlap with current proposal
Role: Staff Investigator

P50CA58236 (Nelson PI/DeWeese Project Leader) SPORE In Prostate Cancer - Project 2: Tissue-Specific Radiation Sensitization of Prostate Cancer by Aptamer-Targeted siRNA Knock-down of DNA Repair Pathways
Time Commitment: .84 calendar (7% effort)
Supporting Agency: NIH/NCI
Grants Officer: Jacquelyn Saval, boudjedaj@mail.nih.gov, 240-276-6312
Address of Funding Agency: NIH, 9000 Rockville Pike; Bethesda, Maryland 20892
Performance Period: 09/25/14-08/31/19
Level of Funding: $128,750 annual direct costs (for Project 2)
Project Goal: The goals of this project are 1) complete a Phase I clinical trial to determine the safety and feasibility of intraprostatically administered radiation sensitization agents for the *in situ* reduction of DNA-PK; 2) to evaluate aptamer-siRNA chimeras as systemically delivered radiation sensitizers in the context of two different models of metastatic disease.
Specific Aims: 1. Clinical evaluation of A10-3-DNA-PK
2. Systemically delivered aptamer-shRNA radiation sensitizing agents
Overlap: No scientific or budgetary overlap

PENDING:

P30CA006973 (Nelson) Regional Oncology Research Center
Time Commitment: 6% (.72 Calendar Months)
Supporting Agency: NCI
Grants Officer: Jacquelyn Saval, boudjedaj@mail.nih.gov, 240-276-6312
Address of Funding Agency: 9000 Rockville Pike; Bethesda, Maryland 20892
Performance Period: 05/01/17-04/30/22
Level of Funding: $4,749,872 annual direct costs
Project's Goal: The major goal of this project is to provide an organizational focus and stimulus to take maximum collective advantage of scientific opportunity and institutional resources aimed toward the ultimate goal of reducing cancer incidence, morbidity, and mortality.
Project's overlap: No scientific or budgetary overlap with current proposal
Role: Staff Investigator